

## LARVAL TREMATODA PARASITISING THE NEW ZEALAND SPECIES OF *POTAMOPYRGUS* (GASTROPODA : HYDROBIIDAE)

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### ABSTRACT

Larval forms of 11 species of Trematoda parasitising the New Zealand species of *Potamopyrgus* (Gastropoda:Hydrobiidae) are described. Fourteen trematodes are now known to use the freshwater species, *P. antipodarum*, as an intermediate host and two of these species have also been found parasitising the brackish water species, *P. estuarinus* and *P. pupoides*. Metacercarial infections were more frequently found than infections by active larval stages and they also produced the highest intrapopulation infection rates. Highest cercarial infection rates were produced by monostome species.

### INTRODUCTION

Interest in the larval forms of trematodes parasitising the freshwater gastropod *Potamopyrgus antipodarum* (Gray) was first shown by workers searching for the molluscan intermediate hosts of the liver fluke, *Fasciola hepatica* L. Hopkirk (1927) believed he had found the larva of *F. hepatica* as well as the cercariae of three other species parasitising *P. antipodarum*, but Macfarlane (1937) showed that this snail was not the intermediate host, although it was infected by "at least 14 other flukes whose adults live in fish, birds and mammals". Later, Macfarlane (1939, 1945, 1951, 1952) described the life histories of three of these species, *Coitocaecum anaspidis* Hickman, *Telogaster opisthorchis* Macfarlane and *Stegadexamene anguillae* Macfarlane, but apart from this nothing has been published.

The observations recorded in the present paper were made during a study of the systematics of the New Zealand species of *Potamopyrgus* (Winterbourn 1970). Although an examination of the parasites was not the principal aim of the work, some interesting information on the larval forms occurring and their incidence was obtained and is presented here.

Although the morphology of the parasites was not examined in detail, brief descriptions and figures are provided to allow future recognition of cercariae and other intermediate stages. Classification of cercariae follows Dawes (1946).

### METHODS

Living snails were squashed between sheets of glass and any larval trematodes present were removed from the snail tissues under a dissecting microscope. Examination of parasites was made with a binocular, compound microscope (up to 400 x magnifi-

cation) and an inverted, phase-contrast microscope at similar magnifications. Methylene blue and methyl green (1% in acetic acid) were used to stain specimens, and photographs were also taken. Measurements were made with a linear eyepiece micrometer. In most cases several specimens of each larval stage were measured after they had been teased from the host's tissues, and in the case of cercariae had died.

Although descriptions of the larval stages of the three species considered by Macfarlane are not repeated here, figures are provided to assist with recognition (Fig. 1).

#### DESCRIPTIONS OF LARVAL TREMATODA

##### MONOSTOME CERCARIAE

###### Cercaria M1 (Fig. 2A, B)

Body length 0.43 - 0.56 mm; width 0.09 - 0.14 mm; tail length 0.11 - 0.15 mm.

Body transparent, granular, with a pair of dark, longitudinal bands following the courses of the main antero-lateral excretory canals; oral sucker inconspicuous; eye spots absent; posterior locomotor pockets (adhesive papillae) weakly developed; posterior angles of body smoothly rounded; excretory vesicle in death circular; tail less than half length of body. Locomotion by a looping action.

###### Redia

Length up to 1.1 mm; width up to 0.29 mm.

Pharynx prominent; active, capable of limited contraction.

###### Metacercarial cysts

Roughly spherical, diameter 0.11 - 0.18 mm. No morphological structures are distinguishable through the cyst wall. Cysts are found attached to the operculum of *P. antipodarum* or to the shell or other cysts close to the aperture. The attached side of the cyst is flattened to fit the curvature of the surface. All snails examined with cysts attached to their shells, contained cercariae. As the incidence of infection within a snail population was never high (Table 1), it seems probable that cercariae habitually encyst on the shell of the snail they have just left.

Molluscan host: *P. antipodarum*

Collection localities: Submerged littoral vegetation, Lake Pupuke, Takapuna; stony stream near Bunnythorpe, Manawatu.

###### Cercaria M2 (Fig. 2C-E)

Body length 0.34 - 0.44 mm; width 0.19 - 0.21 mm; tail length 0.22 - 0.39 mm.

Body opaque, granular, contractile, oval when relaxed, the posterior margin shallowly concave; oral sucker inconspicuous; excretory vesicle circular in death; adhesive papillae prominent; a pair of dark eye spots present anteriorly behind the oral sucker, in some individuals each "eye" appearing to be made up of two adjoining spots; conspicuous band of dark pigment between and around the eyespots; tail simple and contractile. Inactive.

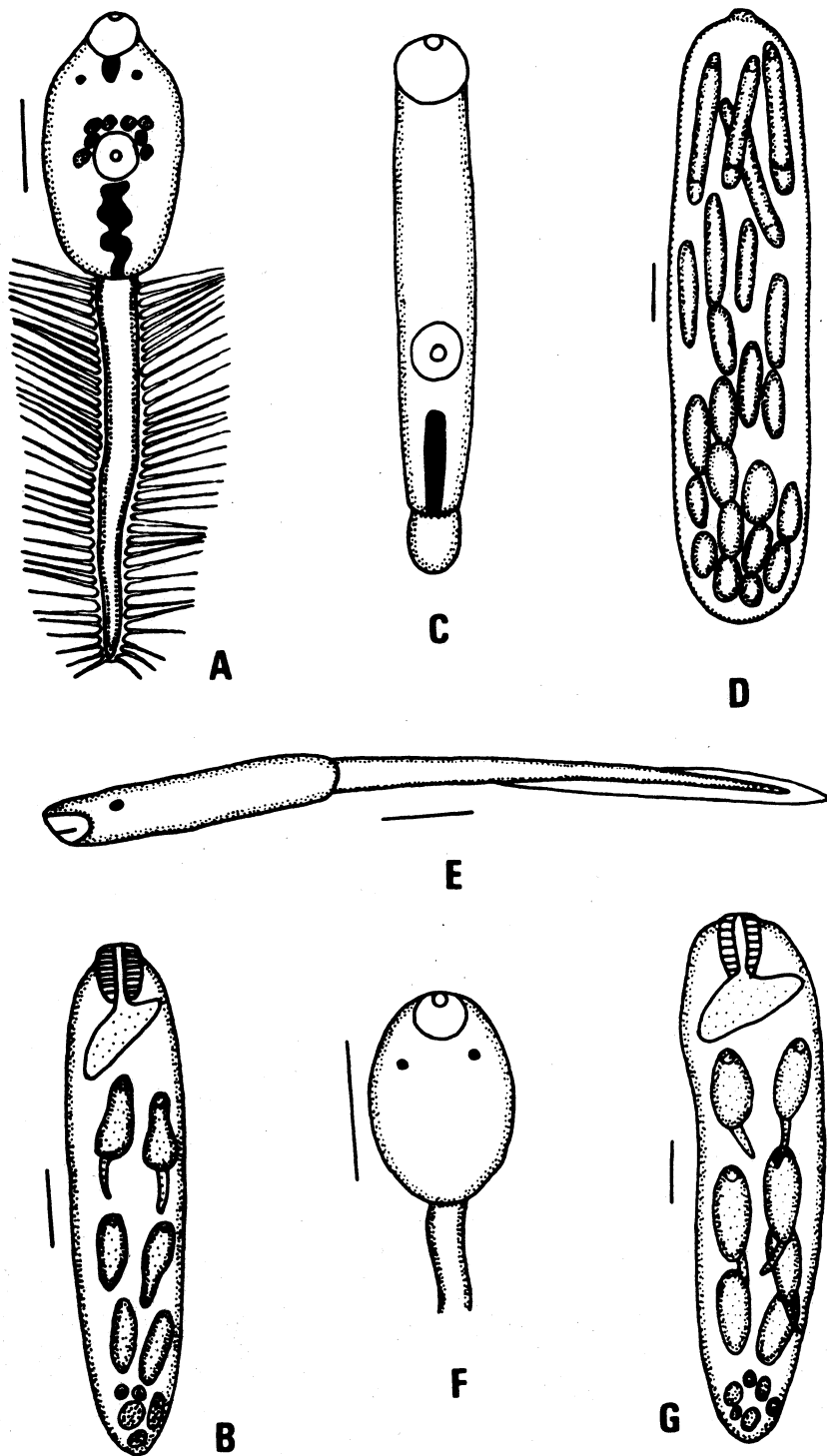


Fig. 1. The trematode species described by Macfarlane. *Stegodexamine anguillae*, A - cercaria, B - redia; *Coitocaecum anaspidis*, C - cercaria, D - sporocyst; *Telogaster opisthorchis*, E, F - cercaria, G - redia. Length of scale lines on this and following figures, 0.1 mm.

### Redia

Similar to that of *Cercaria* M1. Length up to 0.95 mm; width up to 0.38 mm. Containing about 10 cercariae in various stages of development.

### Metacercarial cysts

Diameter 0.17 - 0.23 mm. They occur on the operculum and shell of *Potamopyrgus* spp., often in large numbers, e.g., individual snails from Lake Pukaki had over 300 cysts.

Molluscan hosts: *P. antipodarum* and *P. pupoides* Hutton.

Collection localities: a slow moving, muddy stream near Linton, Manawatu; Hokowhitu Lagoon, Palmerston North; Avon River, Christchurch; marginal vegetation, Lake Pukaki, Canterbury; Makara River, Wellington; littoral vegetation, Lake Tutira, Hawkes Bay; tidal reaches of the Kahao Stream, Porirua Harbour, Wellington.

### *Cercaria* M3 (Fig. 2F, G)

Body length 0.37 - 0.44 mm; width 0.11 - 0.19 mm; tail length 0.33 - 0.55 mm.

Body grey-brown with two opaque, longitudinal bands as in *Cercaria* M1; tail more transparent; three eyespots present, two lateral, one median immediately posterior to the oral sucker; two pronounced adhesive papillae at the posterior body angles; tail with a regular "cellular" appearance.

Cercariae are very active compared with other monostome species found, swimming being accompanied by strong contractions of both body and tail. When attached to a flat surface, the two posterior adhesive papillae are in contact with the substratum and appear to act as substitutes for a ventral sucker.

### Redia

Indistinguishable from those of *Cercariae* M1 and M2.

### Metacercarial cysts

Diameter 0.17 - 0.21 mm. Found attached to shells and opercula of host snails. Encystment was observed on the bottom of a glass dish within minutes of cercariae being teased from a snail host. During encystment the tail thrashed frantically while still attached to the outside of the cyst, and the body of the cercaria rolled continuously within the developing cyst. Initially the three eyespots were clearly visible through the cyst wall but later the wall became more opaque and the internal structures could no longer be distinguished. The cyst wall was built up of two layers, an inner one 7-11  $\mu$ m thick, initially with a golden appearance which was laid down first, and a thicker (15-23  $\mu$ m) outer layer which at first was clear. On completion of encystment the inner layer assumed a grey, somewhat fibrous appearance, and the outer layer appeared more golden when viewed with transmitted light.

Molluscan host: *P. antipodarum*

Collection localities: Hokowhitu Lagoon, Palmerston North; muddy slow-moving tributary to Kahuterawa River, Manawatu.

### *Cercaria* M4 (Fig. 2H, I)

Body length 0.29 - 0.43 mm; width 0.11 - 0.21 mm; tail length 0.29 - 0.56 mm.

Body colourless, opaque; tail more transparent, with a fin

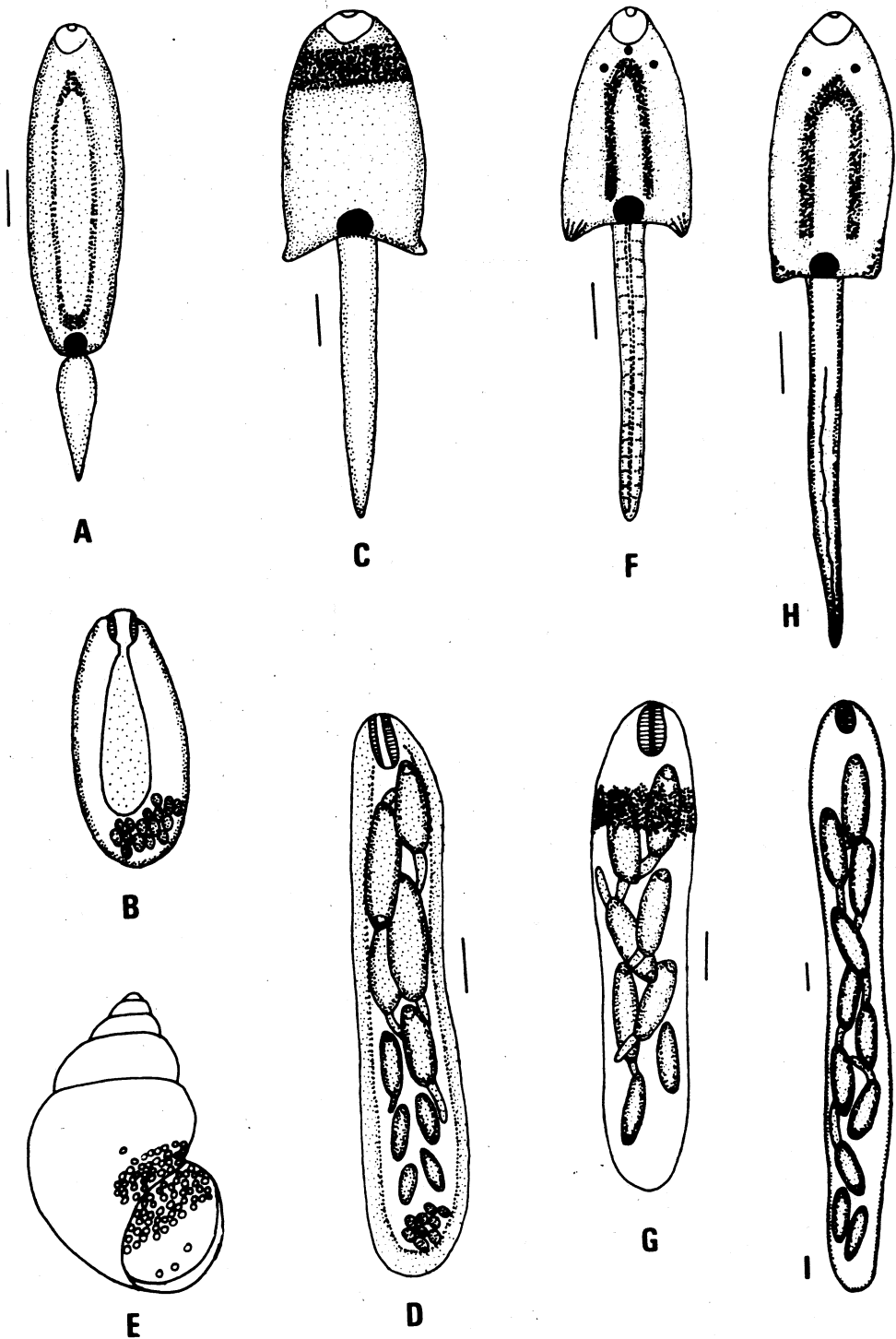


Fig. 2. Monostome cercariae. Cercaria M1, A - cercaria, B - young redia; Cercaria M2, C - cercaria, D - redia, E - metacercarial cysts on *Potamopyrgus* shell and operculum; Cercaria M3, F - cercaria, G - redia; Cercaria M4, H - cercaria, I - redia.

running its full length ventrally and three quarters of its length dorsally; two prominent eyespots present; longitudinal bands of pigment extend the length of the body to the clear, excretory vesicle; oral sucker not easily seen; posterior margins of body "flanged" to form adhesive papillae. The body, but not the tail, exhibits considerable contractility, changing from a roughly circular to a narrow, elongated form. In life the tail is normally about twice as long as the body, but at death it contracts, and approximates the body length. Cercariae swim actively by thrashing movements of the tail which is held vertically above the body.

#### Redia

Length up to 2.0 mm; width up to 0.19 mm.

Translucent with a short, broad pharynx. Inactive. Containing about 11 cercariae in various stages of development.

#### Metacercarial cysts

Indistinguishable from those of *Cercaria* M3. The process of encystment was observed in the laboratory and is also identical to that described for *Cercaria* M3. Found on shells of *Potamopyrgus* spp.

Molluscan hosts: *P. antipodarum* and *P. estuarinus* Winterbourn.

Collection localities: muddy, slow flowing stream near Linton, Manawatu; Avon River, Christchurch; littoral vegetation, Lake Pupuke, Takapuna; tidally exposed mudflats at mouth of Huia River, Manukau Harbour; stony stream near Waitomo, King Country.

#### FURCOCERCOUS CERCARIAE

##### *Cercaria* F1 (Fig. 3A, B)

Body length 0.16 mm; tail stem length 0.08 mm; furcal length 0.12 mm (one specimen measured).

Colourless, translucent, shape rigidly defined; ventral sucker posterior to mid-point of body; excretory vesicle small, V-shaped; tail stem half the length of the body and containing 2 rows of clearly visible caudal bodies; furcae slightly longer than tail stem, tapering towards their apices. An active swimmer.

##### Sporocyst

Length up to 0.44 mm, width up to 0.12 mm.

Molluscan host: *P. antipodarum*

Collection locality: muddy littoral zone, northern end of Lake Wairarapa.

##### *Cercaria* F2 (Fig. 3C, D)

Body length 0.06 - 0.10 mm; width up to 0.04 mm; tail stem length up to 0.15 mm; furcal length up to 0.03 mm.

An apharyngeal, brevifurcate, monostome cercaria. No normal suckers but with a protrusible anterior organ; body with a narrow dorsal fin; tail stem divided into 2 furcae at about three quarters of its length.

##### Sporocyst

Length 0.26 - 0.28 mm; width 0.12 - 0.14 mm.

Molluscan host: *P. antipodarum*

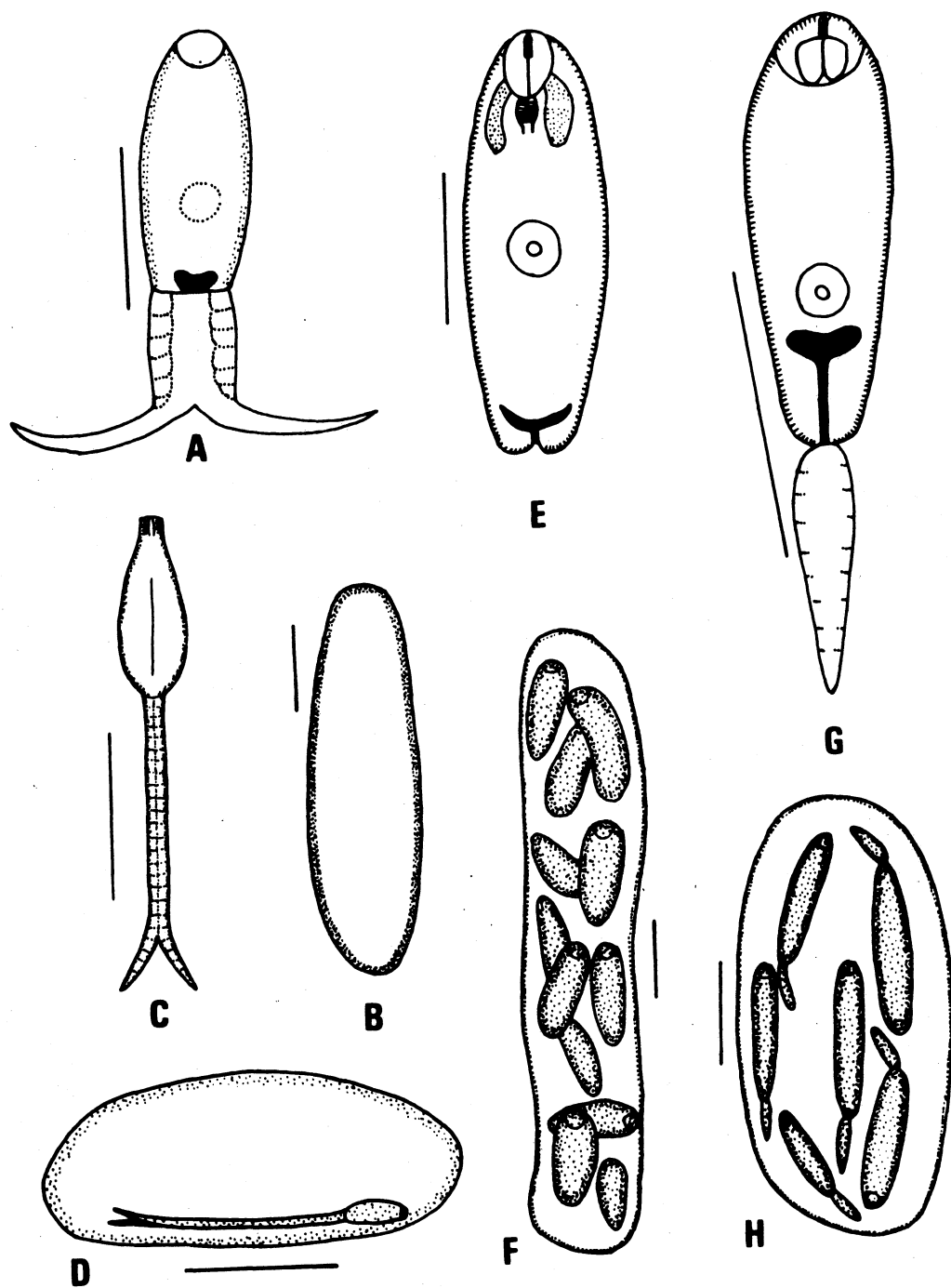


Fig. 3. Furcocercariae, Cercariaea, Xiphidiocercariae. Cercaria F1, A - cercaria, B - sporocyst; Cercaria F2, C - cercaria, D - sporocyst; Cercaria C1, E - cercaria, F - sporocyst; Cercaria X1, G - cercaria, H - sporocyst.

Collection localities: muddy, slow flowing, stream near Linton, Manawatu; littoral vegetation, Lake Tutira, Hawkes Bay.

#### XIPHIDIOCERCARIAE

*Cercaria* X1 (Fig. 3G, H)

Body length 0.15 mm; width 0.06 mm; tail length 0.07 mm (one specimen measured).

Body colourless, opaque; oral sucker larger than ventral sucker, stylet and virgula organ prominent; excretory vesicle V-shaped with a long narrow posterior duct; body surface covered with small spines. Movement by muscular contractions and side to side lashings of the tail.

Sporocyst

Length up to 0.44 mm; width up to 0.19 mm. Containing up to 6 cercariae.

Molluscan host: *P. antipodarum*

Collection locality: littoral vegetation, Lake Tutira, Hawkes Bay.

#### CERCARIAEA

*Cercaria* C1 (Fig. 3E, F)

Body length 0.28 mm; width 0.09 mm (one specimen measured).

Body translucent, white; body surface spinose; oral sucker and stylet prominent; large penetration glands present, one group either side of the oral sucker and extending some distance behind it; pharynx small, inconspicuous; ventral sucker not obvious from above, centrally positioned, almost as large as oral sucker; posterior margin of body drawn in medially to form a shallow V at the excretory pore; excretory vesicle a narrow, 2 branched, Y-shaped structure; tail absent. Cercariae are very active, moving with a looping action.

Sporocyst

Length up to 0.80 mm; width 0.25 mm. Containing about 12 cercariae.

Molluscan host: *P. antipodarum*

Collection locality: stony stream at Waitomo, King Country.

#### GYMNOCEPHALUS CERCARIAE

*Cercaria* G1 (Fig. 4A-D)

Body length 0.20 - 0.26 mm; width 0.07 - 0.09 mm; tail length 0.17 - 0.20 mm.

Body opaque, oval, non-contractile; tail strongly developed with no fin; oral and ventral suckers the same diameter, the latter slightly posterior to the mid point of the body; a pair of eyespots just posterior to the pharynx. Inactive.

Redia

Length up to 1.44 mm; width up to 0.38 mm.

Sac-like, narrowing anteriorly; pharynx small and inconspicuous. Containing up to 11 developing cercariae.

Molluscan host: *P. antipodarum*



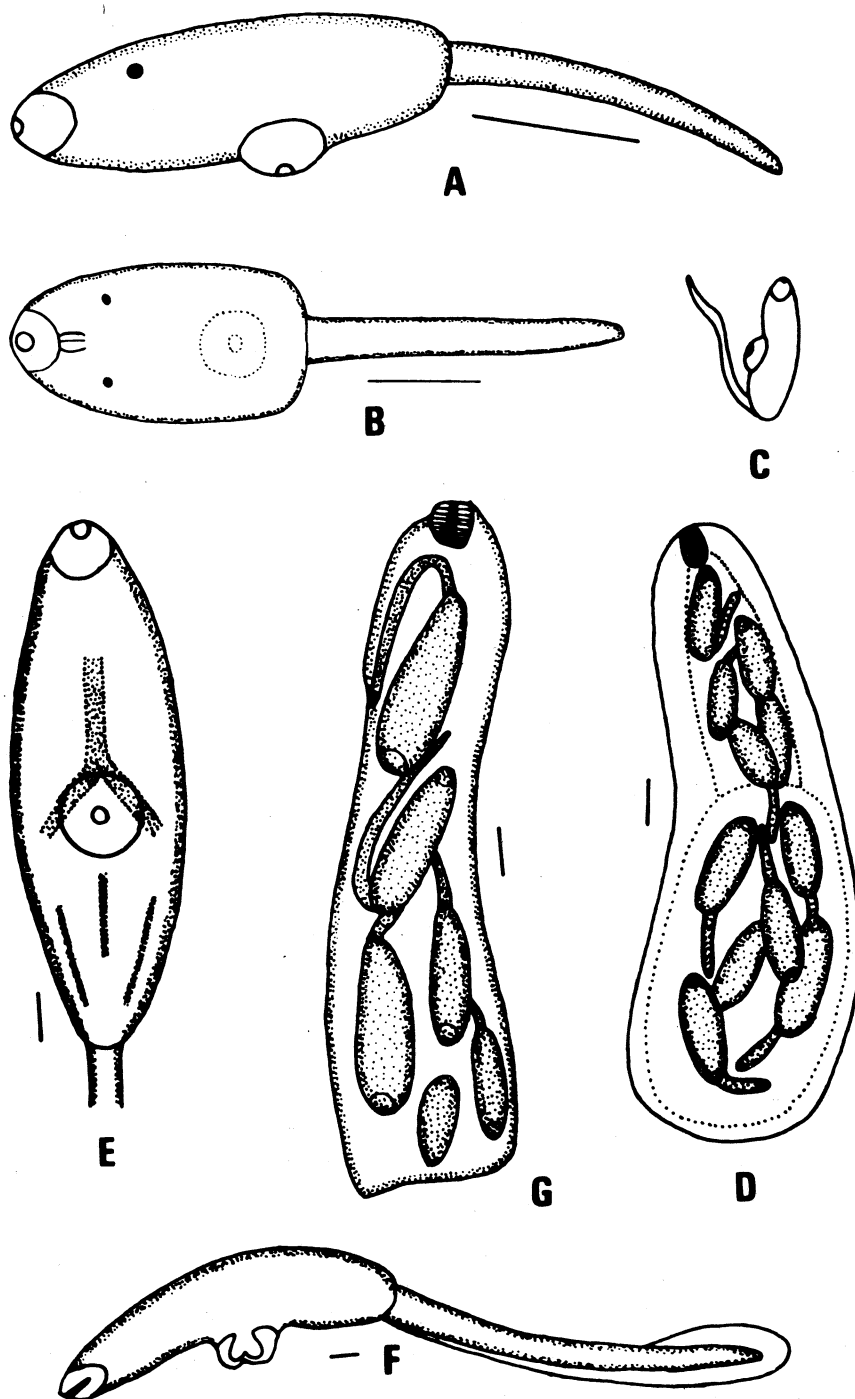


Fig. 4. Gymnocephalous cercariae. Cercaria G1, A, B - cercaria, C - cercaria in swimming position, D - redia; Cercaria G2, E, F - cercaria, G - redia.

TABLE 1. INCIDENCE OF LARVAL TREMATODES INFECTING THREE SPECIES OF POTAMOPYRGUS

Locality	Number in Fig. 5	Month of collection	Sample size	Metacercariae % infection	No. of species	Active stages % infection
<i>P. antipodarum</i>						
L. Pupuke	2	Dec.	100	86	0	0
L. Pukaki	23	Jan.	100	66	1	4
Makara R.	17	Apr.	100	38	1	3
L. Pupuke	2	Aug.	100	28	1	1
Avon R.	19	Jan.	100	27	2	2
L. Tutira	7	Jan.	260	12.8	6	4.6
L. Wahapo	21	Jan.	100	10	0	0
Green L.	4	Jan.	100	10	0	0
Kahuterawa R. (Trib.1)	11	Oct.	100	8	0	0
Hokowhitu Lagoon	8	May	100	8	2	8
Kahuterawa R. (Trib.2)	12	Oct.	84	6	1	1.2
L. Taupo	6	Apr.	35	2.9	2	8.6
Bunynthorpe (stream)	9	Oct.	150	1.3	3	2
L. Paringa	22	Jan.	100	1	1	2
Bunynthorpe (pond)	10	Oct.	200	0	0	0
Linton (stream)	13	Oct.	200	0	4	2.5
L. Wairarapa	14	Jan.	200	0	2	5.5
Waitomo (stream)	5	Dec.	100	0	2	3
<i>P. estuarius</i>						
Huia R.	3	Aug.	200	0	1	0.5
Heathcote R.	20	Jan.	200	0	0	0
Havelock R.	18	Jan.	100	0	0	0
Hutt R.	16	Jan.	50	0	0	0
<i>P. pupoides</i>						
Kahao Stream	15	Oct.	150	0	1	0.7
Wananaki (stream)	1	Dec.	100	0	0	0
Avon R.	19	Jan.	200	0	0	0
Hutt R.	16	Jan.	50	0	0	0
Havelock R.	18	Jan.	50	0	0	0

TABLE 2. MAXIMUM INCIDENCE OF INFECTION OF *P. ANTIPODARUM* BY TREMATODE LARVAE

Species	Maximum % infection
<i>Metacercaria</i> A	86.0
<i>Coitocaecum anaspidis</i>	4.5
<i>Telogaster opisthorchis</i>	0.7
<i>Stegadexamene anguillae</i>	2.0
<i>Cercaria</i> M1	1.15
<i>Cercaria</i> M2	6.0
<i>Cercaria</i> M3	2.0
<i>Cercaria</i> M4	1.2
<i>Cercaria</i> F1	1.0
<i>Cercaria</i> F2	0.8
<i>Cercaria</i> G1	1.2
<i>Cercaria</i> G2	0.4
<i>Cercaria</i> X1	1.2
<i>Cercaria</i> C1	1.0

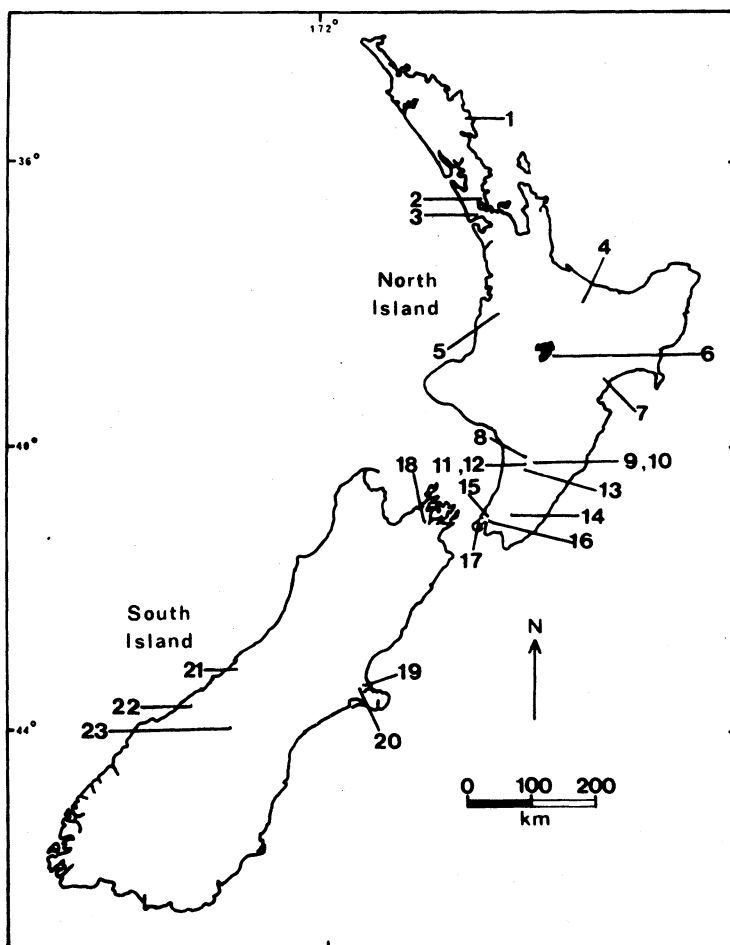


Fig. 5. New Zealand showing collection localities given in Table 1.

Collection localities: muddy, slow flowing stream near Linton, Manawatu; marginal vegetation, Lake Tutira, Hawkes Bay.

Cercaria G2 (Fig. 4F-G)

Body length 1.00 - 1.10 mm; width up to 0.30 mm; tail length 0.75 - 0.87 mm.

The largest cercaria parasitising *Potamopyrgus* species. Body colourless, elliptical; ventral sucker slightly posterior to middle of body, extremely protrusible and larger than oral sucker; bifurcation of the gut clearly visible just anterior to the ventral sucker; tail almost as long as body, a well developed fin running its full length ventrally and along the posterior third dorsally.

Redia

Length up to 1.50 mm; width up to 0.25 mm.

Pharynx prominent. Containing up to 6 developing cercariae. In young rediae, the gut posterior to the pharynx is clearly visible as a simple sac extending almost half the length of the body.

Molluscan host: *P. antipodarum*

Collection locality: marginal vegetation, Lake Tutira, Hawkes Bay.

#### METACERCARIAE

##### Metacercaria A

Cyst spherical, diameter 0.10 - 0.12 mm; thickness of cyst wall 3-6  $\mu$ m. Two dark patches, apparently not structural features of the encysted cercaria are clearly visible through the walls of many viable cysts. Metacercaria with a spinose body surface; anterior sucker 30-36  $\mu$ m diameter; ventral sucker posterior to mid-point of body, 27-36  $\mu$ m diameter; pharynx length 18-24  $\mu$ m.

Infected snails contain hundreds of cysts which pack the spire of the shell and replace much of the digestive gland and gonad, as well as occupying spaces between the viscera.

Molluscan host: *P. antipodarum*

Collection localities: see Table 1.

#### INCIDENCE OF INFECTION OF *POTAMOPYRGUS* SPP.

Infection rates were examined in collections of *P. antipodarum* from 18 localities and in *P. estuarinus* and *P. pupoides* from 6 localities (Table 1). All snails examined had shell heights greater than 3.5 mm (*antipodarum* and *estuarinus*) or greater than 2.0 mm (*pupoides*) and were therefore adult or almost adult individuals.

Little evidence of parasitism was found in *P. estuarinus* or *P. pupoides* and only one infected individual of each species was seen. The parasites concerned (Cercariae M2 and M4) were both monostomes.

Metacercaria A parasitised *P. antipodarum* most frequently (14 out of 18 collections) and also showed the highest intra-population snail infection rates (86% in a collection from Lake

Pukaki). Cercariae were found in 13 collections and in 12 of these monostome species were present. The highest cercarial infection rates (8.0 and 8.6%) were also produced by monostomes (Table 2). No infections of a snail by two or more species of cercariae were found.

#### DISCUSSION

All larval trematodes were found in the region of the digestive gland and gonad of the host snail and infection probably rendered the snail infertile. This was indicated by the observation that no infected individuals of the ovoviviparous *P. antipodarum* contained embryos in their brood pouches, whereas in all cases a high percentage of uninfected snails did so.

Although infections by passive parasite stages (metacercariae) were high in several populations, infections by active stages (rediae, sporocysts, cercariae) were low compared with those recorded in species of Hydrobiidae elsewhere. For example, in populations of the European estuarine hydrobiid, *Hydrobia ulvae* (Pennant), Rothschild (1941) found up to 70% of the snails over 3.75 mm high were parasitised by cercariae and in a Welsh freshwater lake, Probert (1966) found that 45% of *Bithynia tentaculata* (L.) were infected by active stages. Probert also recorded 27% of *B. tentaculata* infected by metacercariae. The numbers of snails found to be parasitised by active stages of individual trematode species in my study were similar to those recorded in New Zealand by Macfarlane (1939) for *Coitocaecum anaspidis* (0.5 - 1.0% in Heathcote River), *Stegadexamene anguillae* (0.7% in Hut River) and *Telogaster opisthorchis* plus *S. anguillae* (2.7% in Heathcote River). The highest snail infection rate recorded by Macfarlane (1952), 15.8% for all trematode species (number and identities not given) in the Heathcote River, however, exceeded the highest cercarial infection rates that I found.

Most species of mollusc that have been examined, act as intermediate host for less than 10 trematode species (Ewers 1964), although there are a number of exceptions including the pulmonate *Lymnaea natalensis* Krauss, the natural host of at least 43 known species of fluke. The use of *P. antipodarum* as first intermediate host by a relatively large number of trematodes (at least 14 species), may have come about because suitable alternative snail hosts are lacking in New Zealand. Probert (1966) found that trematodes which have a prosobranch snail as intermediate host never parasitise pulmonate snails, and *P. antipodarum* is the only prosobranch which is widely distributed in New Zealand freshwaters.

The presence of a large number of host-parasite relationships also indicates that *P. antipodarum* has probably been present in New Zealand freshwaters for a long time and contrasts strikingly with the situation found in Europe and Britain where *Potamopyrgus jenkinsi* (Smith), which may be *P. antipodarum* recently introduced from New Zealand (Winterbourn 1972), is parasite free (Fretter and Graham 1962).

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